# A new polyclad flatworm, *Tytthosoceros inca* (Plathyhelminthes: Polycladida: Cotylea: Pseudocerotidae), from Chilean coastal waters.

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Abstract.—A new polyclad species, *Tytthosoceros inca* is described from Chilean coastal waters. It resembles *Pseudoceros luteus* (Plehn, 1898) Hyman 1953 incerta sedis but differs in having ear-like pseudotentacles, a brown marginal band, and dark brown spots on the dorsal surface of the body. *Tytthosoceros inca* is associated with the colonial ascidians *Ciona intestinalis* and *Pyura chilensis*, living on or near them. This is the first record of the genus from Chilean coastal waters and the second report of the family from cold temperate waters, although this time from the southern hemisphere.

Pseudocerotidae is one of the most populous families of polyclad flatworms (Hyman 1954, Newman & Cannon 1994). Marcus (1950) cited an extensive list of over 70 species, mainly Pseudoceros sensu lato, described from around the world. Hyman (1959a) extended the list to approximately 120, adding many species overlooked by Marcus (1950) and others described by her. Another 60 new species described by Hyman (1959b), Faubel (1984), Prudhoe (1981, 1989), and Newman & Cannon (1994, 1995, 1996a, 1996b) must now be added to the list of species described worldwide, increasing the total number of known species to at least 200.

Most species of the family occur in tropical and subtropical waters of the Indo-Pacific region (Hyman 1954). They have been reported less frecuently from warmer temperate waters around the world, and only one species, *Pseudoceros canadensis* Hyman, 1953 from cold temperate waters around British Columbia, Canada (Hyman 1953). However, this species is considered incerta sedis due to its incomplete description (Faubel, 1984; Newman & Cannon, 1984).

At present, the following 14 pseudocerotid genera are recognized; Thyzanozoon Grube, 1840, Acanthozoon Collingwood, 1876, Pseudoceros Lang, 1884, Yungia Lang, 1884, Nymphozoon Hyman, 1959, Cryptoceros Faubel, 1984, Cryptobiceros Faubel, 1984, Monobiceros Faubel, 1984, Pseudobiceros Faubel, 1984, Parapseudoceros Prudhoe, 1989, Bulaceros Newman & Cannon 1996, Tytthosoceros Newman & Cannon 1996, Maiazoon Newman & Cannon 1996, and Phrikoceros Newman & Cannon 1996. They can be easily distiguished by details of the male and female reproductive systems (presence or absence of prostate and seminal vesicle, and the number of gonopores and complexes), the alimentary system (presence or absence of anal pores), and by external characters, such as the shape of the body, pseudotentacles, pharynx and pharyngeal lobes, and the arrangement of the cerebral and pseudotentacular eyes (Newman & Cannon 1994, 1995, 1996a, 1996b).

Nevertheless, species recognition and differentiation within each genus presents problems in this group, like in many others that lack solid structures. Marcus (1950), Hyman (1954, 1959a), Faubel (1984), Prudhoe (1989), and Newman & Cannon (1994) have commented extensively on the subject. Authors based species identification primarily on body colour pattern (Hyman 1954, 1959a, Prudhoe 1989, Newman & Cannon 1994). Aspects of the male copulatory apparatus, such as the relationship between size of the seminal and prostatic vesicles, cannot be used, due to the extreme similarity of the complex within each genus and considering that some features can change during development or can be deformed by histological procedures (see Prudhoe 1989, Newman & Cannon 1994).

In Chile, the only study on polyclad flatworms is that of Marcus (1954), based on specimens collected by the Lund's University Chile Expedition. No other research has been conducted and Pseudocerotidae species have not been reported from the country to date.

During exploratory diving in northern Chile we observed a pseudocerotid polyclad which belongs to the recently described genus, *Tytthosoceros* Newman & Cannon, 1996. This is the first record of the genus from Chilean coastal waters (Marcus 1951) and the second report of the family that we have noticed from cold temperate waters, in this case in the southern hemisphere.

# Materials and Methods

*Tytthosoceros inca* specimens were collected from lantern nets with *Argopecten purpuratus* (Lamarck, 1819) (Pectinidae) located along a dock at the Universidad Católica del Norte, in Bahía La Herradura, Coquimbo (29°58'S, 71°22'W), Chile. Flatworms were observed on the fouling of the lanterns, typically in association with the ascidian *Ciona intestinalis* (Linnaeus, 1767).

Four specimens were mounted *in toto* after fixation in Bouin-Hollande, stained with Delafield's hematoxylin, and cleared with xylol.

For histology, specimens were fixed in Bouin-Hollande, embedded in paraffin, serial sectioned at seven micrometers, and stained with Harris hematoxylin and eosin. Photographs were taken using a Nikon Biophot microscope and drawings of the male and female copulatory apparatus were made.

The specimens selected as type material were fixed in Bouin-Hollande, preserved in 70% ethanol, and deposited in the Museo Nacional de Historia Natural, Santiago, Chile (MNHNC) and in the Sala de Sistemática, Departamento de Ecología, Pontificia Universidad Católica de Chile, Santiago (SSUC).

## Results and Discussion

# Tytthosoceros inca, new species Figs. 1-3

*Material examined.*—North Chile: Bahía La Herradura, Coquimbo, 2 m, D. Véliz & L. M. Pardo coll., from scallop lantern nets associated with the ascidian *Ciona intestinalis*, 26 Feb 1996, holotype (MNHNC-PL. 11143), 2 paratypes (MNHNC-PL. 11144 and 11145), and 1 specimen (SSUC 6765); Bahía La Herradura, from lantern nets, 1 m, J. A. Baeza coll., Oct 1994, 7 specimens (4 cleared and 3 sectioned); Bahía La Herradura, associated with *Pyura chilensis* Molina, 1782 (Tunicata), 0.5 m, R. Acuña, P. Romero and J. C. Villarroel coll., Apr 1993, 7 specimens.

*Description.*—External morphology: Mature living specimens are extremely soft and thin, reaching a maximum length and width of 11 cm and 6 cm, respectively. The body is ovoid in small animals, becoming elongated-ovoid in worms over 5 cm long. The body margins are broadly folded, caudal and anterior margins are rounded. The dorsal surface is smooth. A pronounced middorsal ridge extends along the animal, ending blindly some distance before the posterior margin (Figs. 1, 2A). Marginal pseudotentacles are weakly developed and ear-like, formed as is usual by the upfolding



Fig. 1. *Tytthosoceros inca*, new species. Photograph of living organism, dorsal view. Scale bar in centimeters.

of the anterior margin (Figs. 2B, C). Each pseudotentacle has a pronounced auricular groove ventrally (Fig. 2C).

On living animals, the mouth opens in the mid-ventral line, in middle of the pharynx. The pharynx begins on the second ninth of the animal, occupying about one fourth or one fifth of body length. The male gonopore opens beneath the posterior end of the pharynx over a promontory. The female pore is located close behind it, about one half the distance between the mouth and the male apperture. The ventral sucker is conspicuous and circular, lying anterior to the middle of the body. The distance between the sucker and female apperture is about three or four times the distance between the male and female pores (Fig. 2A).

Color: The ground color of the dorsal

surface on living animals is brownish-yellow with a narrow marginal white stripe and a broader border-band of pale brown around the body, except on the pseudotentacles (Fig. 1). The dorsal surface also shows many dark brown dots with no special arrangement, decreasing in size to the margins but increasing in number. A narrow black stripe extends along the center of middorsal ridge. This stripe begins at the proximal end of the pharynx, immediately behind cerebral eyespot, ending somewhat before the ridge (Figs. 1, 2B). Only the tip presents the white marginal stripe on the pseudotentacles. Along the inner border of each pseudotentacle, a dark brown stripe exists, narrowing towards the tips and widening at the pseudotentacular bases. Both stripes are not connected with the middorsal one. Another pair of dark brown stripes extends marginally between pseudotentacular bases (Fig. 2B). Ventrally, the color is brownish-yellow with a marginal white stripe and pale brown border-band around the body.

Eyes: Ocelli of chalice type. Cerebral eyespot present as a single oval cluster in small animals or horseshoe shaped in worms over 6 cm length (Fig. 2B). In all animals over 6 cm length, a pair of precerebral eyes lies deep in the parenchyma some distance before the cerebral cluster. Few pseudotentacular eyes extend along the dorsal surface of pseudotentacles. There are also eyes scattered dorsally, between pseudotentacular bases (Fig. 2B). Ventrally, pseudotentacular eyes are more numerous, extending as two irregular clusters along the margins of pseudotentacles, ending as a pair of inverted triangles between the pseudotentacular bases (Fig. 2C).

Digestive system: Ruffled pharynx deeply folded and contained inside the pharyngeal chamber formed by the anterior portion of the central ridge. Behind the pharynx chamber, the ridge contains the main intestinal branch that extends to the posterior end of the body, giving rise along its course to numerous branches that divide to

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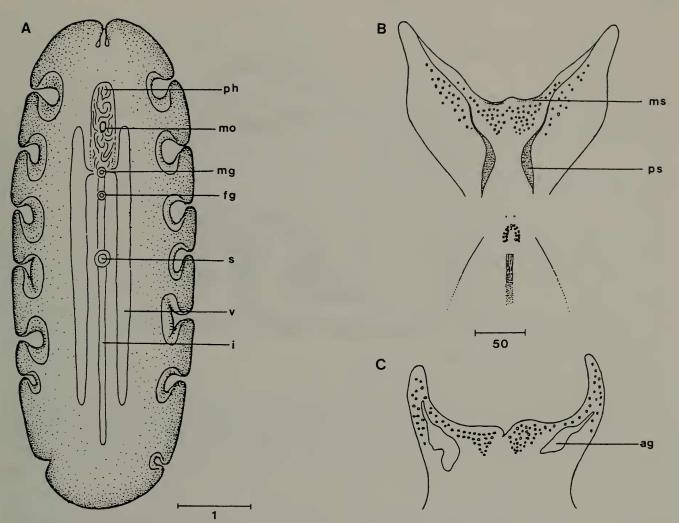


Fig. 2. *Tytthosoceros inca*, new species. A, ventral view of body; B, anterior portion of body in dorsal view, showing pseudotentacle morphology, eye arrangement and color pattern; C, pseudotentacles, ventral view (MNHNC-PL. 11143). Scale bars in centimeters (A) and micrometers (B and C). Abbreviations: ag = auricular groove, fg = female gonopore, i = main intestine, mg = male gonopore, ms = marginal inter-pseudotentacular stripe, mo = mouth, ph = pharynx, ps = dorsal pseudotentacular stripe, s = sucker, v = vas deferens.

form a dense anastomosed network. It seems that the main intestine does not extend further over the pharynx, but some diverticules can be found.

Copulatory apparatus: The vas deferens form a prominent network extending forward and backward, running along each side of the main intestine. A single male copulatory complex exists (Fig. 3). Two sperm ducts connect spermiductal vesicles with the posterolateral extremes of an ovalelongated seminal vesicle. At its proximal end it narrows to form a sinuous ejaculatory duct that joins the base of a conical penis papilla. At the same location on papilla the ovoid prostate or prostatic vesicle is connected by a short prostatic duct. The seminal vesicle is about three times the size of the prostate. The penis papilla is armed with a short and wide cuticular stylet both surrounded by a penis sheath. The length: width ratio of the stylet is 1:3.5. The male antrum is narrow and deep.

The female gonopore leads into a deep female antrum that opens into a laterally expanded cement pounch. On all sectioned specimens, the vagina was never observed connected with any cement gland. From the cement pounch the vagina ascends, surrounded by the cement gland. The vagina then curves and receives the uteri (Fig. 3).

Geographical distribution.—Known from Bahía La Herradura (29°58'S,

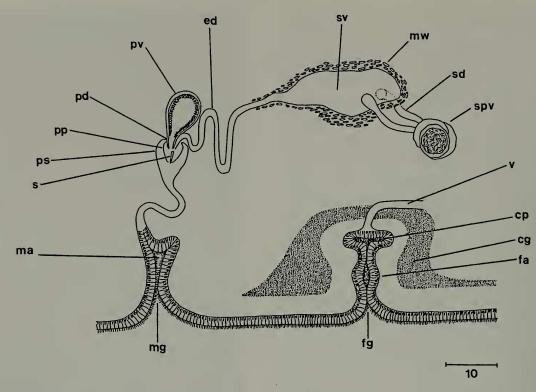


Fig. 3. *Tytthosoceros inca*, new species. Male and female copulatory apparatus, sagital view. Scale bar in micrometers. Abbreviations: cg = cement gland, cp = cement pounch, ed = ejaculatory duct, fa = female antrum, fg = female gonopore, ma = male antrum, mg = male gonopore, mw = muscular wall, pd = prostatic duct, pp = penis papilla, ps = penis sheath, pv = prostatic vesicle, s = stylet, sd = sperm ducts, spv = spermiductal vesicles, sv = seminal vesicle, v = vagina.

71°22'W), Guanaqueros Bay  $(30^{\circ}04'S, 71^{\circ}23'W)$ , and Tongoy Bay  $(30^{\circ}14'S, 71^{\circ}28'W)$ , north Chile.

Taxonomical remarks.—The presence of small ear-like pseudotentacles, the arrangement of dorsal eyes, and the short and wide penial stylet allow us to classify the studied type specimens as Tytthosoceros, a genus recently erected by Newman & Cannon (1996a). This genus has a single reproductive system, but on its overall morphology it resembles Pseudobiceros Faubel, 1984 wich possesses paired male systems (Newman & Cannon 1994). Studied specimens can be distinguished from Bulaceros Newman & Cannon 1996, which has two cerebral eye clusters (not a horseshoe shaped cerebral eyespot), a flat body shape (without a pronounced ridge) and a weakly sclerotised, small, and narrow penial stylet. Moreover, the pronounced central ridge of the body, the ear-like tentacles, and the dorsal eye arrangement of type specimens distinguish T. inca from Pseudoceros, which has a flat body shape, simple pseudotentacles, and dorsal eyes disposed in two to three scattered lines across the anterior pseudotentacular rim (Newman & Cannon 1994).

With regard to the overall body shape and the colour pattern (ground colour of the body and medial black stripe), *Tytthosocer*os inca resembles only one species, *Pseu*doceros luteus (Plehn, 1898) previously reported from Monterrey Bay and Corona del Mar, California coast (Hyman 1953). The latter is considered as incerta sedis, since Newman & Cannon (1994) did not considered it as a member of *Pseudoceros* sensu stricto when they reviewed the taxonomy of the group.

Both species can be distinguished by details of pseudotentacular morphology and colour pattern. While in *P. luteus* the tentacles are broad and flap-like (Marcus 1953), in *T. inca* they are weakly developed and ear-like, with respect to body marks, *P. luteus* lacks the pale brown border-band and

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the dark brown spots on the dorsal surface observed on the body of *T. inca.* Also, the middorsal stripe in *P. luteus* forks before the cerebral eyes and continues along the anterior margin of the body to connect with a pair of large oval spots (Plehn 1898, Hyman 1953), while in *T. inca* this stripe ends immediately behind the cerebral eyes and is not associated with any other markings.

All features mentioned above have been considered as important diagnostic characters by Marcus (1950), Hyman (1954), Prudhoe (1984), and Newman & Cannon (1994, 1995, 1996a, b), allowing us to differentiate *Tytthosoceros inca* from *Pseudoceros luteus* and to describe the former as a new species.

Hyman (1953) also reports a mottled gray color variant of *Pseudoceros luteus*. In our opinion, it is probably another species due to colour pattern differences with *P. luteus*. It is also appropriate to compare the new species with this color variant. *P. luteus* and the color variant of *P. luteus* present the same morphological differences with respect to *T. inca*. Concerning body marks, the color variant can be distinguished from *T. inca* because it lacks the middorsal black stripe and the pale brown border-band found in the new species.

The main diagnostic characters that differ between this new species and other Tytthosoceros are; the pronounced male antrum, the extremely short prostatic duct and the long ejaculatory duct, the long and narrow stylet (largest in T. inca, length:width ratio 1:1.35, compared to 1:2 for T. lizardensis Newman & Cannon 1996 and T. nocturnus Newman & Cannon 1996), and the presence of a pair of pre-cerebral eyes lying deep in the parenchyma in all animals over 6 cm length (not present in others congeners). Due to these differences, it is possible that the present specimens warrant a new genus (Newman, pers. comm.). However, due to the lack of complete morphological and anatomical information on previously described pseudocerotids, we considered our specimens as members of Tytthosocer*os.* We believe that more information is needed on several species in order to erect a new genus.

*Etymology.*—The name *inca* refers to the Incan civilization that occupied the northern and north-central area of Chile during the Pre-Hispanic age.

*Ecological remarks.*—This species has also been observed in Bahía La Herradura and Tongoy in shallow waters, living close to or on the colonial ascidian *Pyura chilensis.* It probably feeds directly on the ascidians or may prey on the bivalves when inhabiting lantern nets. The former feeding habit has been previously described for other species of the family (Hyman 1951, Newman & Cannon 1994). *Tytthosoceros inca* also can be observed in dense aggregations on the bottom or swimming towards the surface with undulating movements.

This is the first record of the genus *Tyt*thosoceros from coastal Chilean waters. Since until now only one species of Pseudocerotidae has been cited for cold temperate waters, this appears to be the second record for the family, although this time from the southern hemisphere.

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